CHAPTER V

CONCLUSIONS AND SUGGESTIONS FOR FUTURE WORK

Ester analytes with different substitution patterns could be successfully prepared and separated with either heptakis (2,3-di-*O*-methyl-6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin (or BSiMe) or heptakis (2,3-di-*O*-acetyl-6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin (or BSiAc), or both of them. Substitution of C2- and C3-methoxy groups with acetoxy groups on cyclodextrin molecule changes enantioselectivity for almost all analytes. The separation of most of selected chiral esters was attained from BSiAc column. Excellent enantioselectivity of this phase is probably ascribed to the polar interactions and hydrogen bonding between acetoxy groups on BSiAc molecule and substituents on ester analytes. However, in some cases, the separation of enantiomers may be hampered by steric hindrance of these acetoxy groups, which in turn led to the loss of enantioselectivity.

In order to acquire more information about the influence of analyte structure on the chiral separation on BSiMe and BSiAc phases, thermodynamic parameters of these esters were systematically investigated using two different methods, *van't Hoff approach* and *Schurig approach*.

From thermodynamic data obtained, as a rule, the ΔH and ΔS values on two chiral columns are greater than those on nonchiral polysiloxane (higher negative values), which indicates stronger interaction and more interaction sites between analytes and chiral phases. This is likely due to the increased interaction between analytes and cyclodextrin derivatives, resulting in longer retention on the chiral columns. Besides, the interaction strength (ΔH) does not correlate with the discrimination of enantiomers (Δ(ΔH)). Ester analytes having the strongest interaction with stationary phase does not always exhibit the greatest enantioselectivity.

Apparently, the interaction and enantioselectivities of esters on these two chiral phases depend on ester chain length, position of substituent, and type of substituent. By *van't Hoff approach*, lengthening ester chain generally increases the intensity of interaction but decreases enantioselectivity of analytes towards stationary

phases. Changing the position of substituent or chiral center also affect the chiral recognition. With regard to type of substituent, there are many contributions from substituent on separations and in some cases the combination of these factors was assumed. It is observed that the presence of polar substituents such as hydroxy, methoxy, and phenoxy groups offers additional polar interaction with stationary phases and consequently affects the separation. In addition to the polarity of substituent, enantioselectivity depends on the size of substituents as well. In addition to the analyte structure, the enantioseparation is influenced by the substituents on CD molecules. Esters with small or polar substituents could be better separated on BSiAc than BSiMe due to polarity and steric effect of BSiAc phase.

Nonetheless, one drawback exists in *van't Hoff approach*. Even though the concentration of the cyclodextrin derivatives in OV-1701 polysiloxane is the same, the amount of OV-1701 matrix is different for both of the chiral columns because of the unequal molecular weight of BSiMe and BSiAc. Consequently, ΔH and ΔS values resulted from the cyclodextrin derivative and polysiloxane cannot be subtracted directly from each other. As a result, the comparison of ΔH and ΔS values between two chiral columns should be considered only a guideline for selecting an appropriate stationary phase for each analyte or a trend to compare the effect of analyte structure on separation on each stationary phase.

In order to solve the problem of nonchiral polysiloxane, *Schurig approach* was used for the determination of thermodynamic values. The results showed that there were large differences in thermodynamic parameters calculated by *van't Hoff approach* and *Schurig approach*. However, *van't Hoff approach* provided thermodynamic results in better agreement with chromatographic results than *Schurig approach* did. It was assumed that thermodynamic data obtained from *Schurig approach* were erroneous, probably due to nonlinearity of the plots of ln R' versus $1/T$, and too small data points used for the calculation of enthalpy and entropy values.

Nevertheless, the conclusions of this model study about the effect of ester structure on enantioseparation cannot be generalized from the results obtained in this study due to the limited number of groups of analytes. Therefore, a further study should cover wider range of esters with various substitution patterns. Moreover,

molecular modelling experiments should be performed in order to prove the aforementioned assumption about analyte-stationary phase interaction binding and to obtain more information about chiral separation mechanism.